(12) UK Patent Application (19) GB (11) 2 174 904 A

(43) Application published 19 Nov 1986

(21) Application No 8610588

(22) Date of filing 30 Apr 1986

(30) Priority data

(31) 1826/85 (32) 30 Apr 1985 (33) CH

(71) Applicant
Seuref A. G. (Liechtenstein),
Austrasse 27, FL-9490 Vaduz, Liechtenstein

(72) Inventor
Pierre-Noel Brasey

(74) Agent and/or Address for Service
Batchellor, Kirk & Eyles, 2, Pear Tree Court, Farringdon Road, London EC1R 0DS

(51) INT CL⁴
A61K 31/12

(52) Domestic classification (Edition H):
A5B 170 180 23X 23Y 301 303 30X 30Y 426 42Y 482 483 48Y 502 503 50Y 550 55Y 576 57Y 586 58Y 616 61Y 650 65Y J
U1S 2415 A5B

(56) Documents cited
GB A 2137088

(58) Field of search
A5B
Selected US specifications from IPC sub-class A61K

(54) **Vasculoprotecting compositions**

(57) The compositions contain an ubiquinone compound and one or more compounds of known vasculoprotecting activity, e.g. of flavanoid, heparinoid, terpenic or glycosidic structure, such as escin, troxerutin, asiaticoside, heparin, delphinidin and tribenoside. The ubiquinone is preferably Coenzyme Q10.

GB 2 174 904 A

SPECIFICATION

Vasculoprotecting pharmaceutical compositions

This invention relates to pharmaceutical compositions for vascular therapy.

Among the vasculoprotecting agents, particularly known are flavonoids, whose effectiveness as capillary-protecting agents, antiedemics and cicatrizants in vasal diseases has been widely described in literature (Bohr D.F. - J. Pharmacol. Exp. Ther., 97-243-1949; Gugler R. - Ges. Arch. Exp. Pathol. Pharm. - 274-R45-1972; Haeger K. - Zbl. Phlebol., 6-526-1967; Rozkoch K. - Europ. J. Clin. Pharmacol., 3-243-1971; Allen S. - Practioner, 205 - 221-1970).

Similar activities were disclosed for heparin and heparinoids (Witte S., Thromb. Diath. Hemorrhag., 2-146-1958; Jacques L. B. - Ann. N. Y. Acad. Sci., 115-781-1969; Nicolaides A.N. - Lancet, 2-890-1972).

All the flavonoids, at the vasal level, act to regulate the entry of calcium into the cells (Ludwig O. - Med. Welt, 16-1181-1942) and to inhibit any increased effect of lysosomial enzymes, related to pathologic conditions (Niches P. - Angiologica, 8-297-1971).

Moreover, it should be pointed out that the synthesis of glycosaminoglycanes, which are among the main components of vasal wall and control its hydropexic action as well as ion passage, takes place mainly by activation of Golgi apparatus, whose function is conditioned by the availability of respiratory chain, related to Coenzyme Q_{10} (Balasz E. - Arthritis-Arthrose Verlag Haan Huber, Bern Wien, 46-1971; Crane F. L. - "Biomedical and Clinical Aspects of Coenzyme Q", Folkers K. Ed., Vol. 1, Elsevier Amsterdam, 1977, p. 3; Lenaz G. - Drugs Exptl. Clin. Res., 10-481-1984).

A deficiency in oxygen diffusion and utilization, as well as tissular hypoxic conditions, have been also indicated as causes of vascular diseases and atherosclerosis (Robertson A.L. - Progr. Biochem. Pharmacol., Vol. 4, p. 305, Karger, Basel, 1968; Zempleny T. - Symposia Angiologica Santoriana - 4 Int. Symp., Fribourg-Nylon 1972 - Angiologica, 9-429-1972).

Coenzyme Q_{10} is known to control the transport of mytochondrial electrons, and consequently to play a role in oxygen utilization and tissue metabolism, being tissular and cellular energetic processes related to hydrogenion transport (Morton R.A. - Nature, 182-1764-1958; Gale P.H. - Arch. Biochem. Biophys., 93-211-1961).

In fact, Coenzyme Q_{10} avoids the damages caused by cellular lypoperoxydation, intracellular calcium increase and lysosomial enzyme release, which markedly affect normal cellular and tissular vasal functioning (Lehedev A.V. - J. Mol. Cell. Cardiol., 14 (Suppl. 3) 99-1982; Littarru G.P. - Drugs Exptl. Clin. Res., 10-491-1984).

Now it has surprisingly been found that ubiquinone compounds, particularly Coenzyme Q_{10}, synergetically enhance the activity of known vasculoprotecting agents, particularly chalones, aurones, flavones, flavanones, flavanols, flavanonols, flavanediols, leukoanthocyanidines, cumarins, hesperidins, catechins, anthocyanidines, natural or semisynthetic derivatives thereof, or vegetal extracts containing them.

According to the present invention we provide pharmaceutical compositions containing as the active ingredient an ubiquinone compound together with known compounds suitable for vascular therapy.

Examples of said vasculoprotecting compounds are those having flavonoid, terpenic, glycosidic and heparinoid structures, such as escin, rutin, troxerutin, diosmin, asiaticoside, hesperidin, tribenoside, etc.

We have found that the components of the compositions according to the invention display a synergetic action on the regulation of vascular activity and permeability.

Moreover, said compositions prevent trophic and metabolic tissular changes as well as those from anoxic conditions, due to impaired vascularization.

By means of the combination of the present invention, a marked increased in therapeutic action is obtained, in comparison with the one of the single components, without toxic or side-effects.

The pharmaco-toxycologic tests carried out will be hereinafter described, in order to illustrate the advantages of the compositions of the present invention.

Coenzyme Q_{10} is known to have a poor toxicity, its DL_{50} in rats and mice being respectively higher than 500 and 250 mg/kg by intraperitoneal administration, and higher than 4 g/kg by subcutaneous administration.

Flavonoids and heparinoids are also known to be poorly toxic, and the combination thereof with Coenzyme Q_{10} shows no changes in the toxicity values of the components. 1:1 Combination of Coenzyme Q_{10} and anthocyanine, hydroxyethylrutoside or catechin, showed DL_{50} values higher than 1 g/kg in both rats and mice, by oral administration.

Also the chronic toxicity tests carried out in the rat by oral daily administration of the above combinations in 1:1 ratio for 30 consecutive days showed no pathologic changes in the body weight and in nemathological and biochemical parameters.

On the contrary, the above combinations proved to be surprisingly effective in different pharmacological tests, such as wound-healing time in experimental wounds in the rabbit, rupture resistance of cicatrices in the mouse, antiedemic activity in the rat, activity on capillary permeability and ergotamine arteritis in the rat.

In all the above tests a marked synergism between Coenzyme Q_{10} and the other vasoprotecting compounds was evidenced, the obtained results being surprisingly more favourable than the ones obtained using the single components.

The tests were carried out combining Coenzyme Q_{10} with O-(β-hydroxyethylrutoside), escin (a saponin from Esculus hippocastanum), tribenoside (ethyl 3,5,6-tri-O-benzyl-δ-glycofuronoside), delphinidin

(anthocyanidin), and total extract of Centella asiatica (asiaticoside).

The tests on wound healing activity of the compositions of the invention were carried out on cutaneous wounds in the rat orally treated with prednisolone, according to the procedure of J.J. Morton (Morton J.J.P., Malone M.H. - Arch. Int. Pharmacodyn., 196-117-1972).

The cicatrization of the cutis of the back of the rat previously scarified was delayed by oral treatment with 0.25 mg/kg of prednisolone.

The test composition was applied on the wound, after appropriate suspension at different concentrations.

The area of the wound on the third day of treatment was measured: the results reported in the following Table 1 evidence a marked synergetic action of the compositions of the invention.

TABLE 1

Compound	*Dose mg/cm² cutis*	*% Reduction*
A	100	22
B	10	15
C	50	11
D	25	18
E	25	12
A + B	100 + 10	65
A + C	100 + 50	40
A + D	100 + 25	56
A + E	100 + 25	63

A = Coenzyme Q_{10}
B = Delphinidin
C = Escin
D = Tribenoside
E = Asiaticoside.

Oral administration of the compositions of the invention also surprisingly increased the rupture resistance of scars induced in the mouse by incision of cutis of the back. The treatment was continued during 3 consecutive days after surgical suturation of the wound, after this period the cutis portion containing the scar was removed, one end thereof was fixed to a support and the other one was subjected to continuous traction (110 g/minute). The rupture strength results are reported in Table 2 hereinbelow.

TABLE 2

Compound	*Dose mg/kg (per os)*	*% Rupture resistance (in grams)*
A	25	19
B	100	21
C	25	16
D	50	15
E	25	13
A + B	25 + 100	75
A + C	25 + 25	82
A + D	25 + 50	80
A + E	25 + 25	95

A, B, C, D, E = *vide* Table 1.

The capillary resistance tests were carried out according to the procedure of Charlier R. (Charlier R., Hosslet A., Colot M. - Arch. Int. Physiol. Biochem., 71-1-1963), by oral treatment for 3 consecutive days of rats, previously fed for 2 weeks with a diet lacking in vitamin P.

The increase of capillary resistance was measured by means of a mercury vacuometer.

The results are reported in following Table 3.

TABLE 3

Compound	*Dose mg/kg (per os)*	*Capillary resistance % increase*
A	25	2.5
B	100	12.2
C	25	9.5
D	50	12.9
F	50	11.5
A + B	25 + 100	35.5
A + C	25 + 25	24.3
A + D	25 + 50	32.4
A + F	25 + 50	25.5

A to D = *vide* Table 1.

F = Hydroxyethylrutoside.

Also in this test a marked synergetic action was evidenced.

The pharmaceutical compositions of the invention may be prepared according to conventional procedures

of pharmaceutical technique, using pharmaceutically acceptable carriers or diluents

Non-limiting examples of suitable compositions of the invention are reported hereinbelow.

Tablets

- Coenzyme Q_{10} 10 mg + escin 20 mg
- Coenzyme Q_{10} 50 mg + escin 20 mg
- Coenzyme Q_{10} 10 mg + hydroxyethylrutoside 300 mg
- Coenzyme Q_{10} 50 mg + hydroxyethylrutoside 500 mg
- Coenzyme Q_{10} 10 mg + asiaticoside 10 mg
- Coenzyme Q_{10} 50 mg + asiaticoside 10 mg
- Coenzyme Q_{10} 10 mg + tribenoside 400 mg
- Coenzyme Q_{10} 50 mg + tribenoside 500 mg
- Coenzyme Q_{10} 10 mg + delphinidin 100 mg
- Coenzyme Q_{10} 50 mg + delphinidin 200 mg.

Sugar-coated pills

- Coenzyme Q_{10} 10 mg + escin 20 mg
- Coenzyme Q_{10} 50 mg + hydroxyethylrutoside 500 mg
- Coenzyme Q_{10} 10 mg + tribenoside 500 mg
- Coenzyme Q_{10} 50 mg + tribenoside 500 mg

Capsules

- Coenzyme Q_{10} 10 mg + escin 20 mg
- Coenzyme Q_{10} 10 mg + tribenoside 400 mg
- Coenzyme Q_{10} 50 mg + tribenoside 500 mg

Creams

- Coenzyme Q_{10} 1% + escin 1%
- Coenzyme Q_{10} 1% + hydroxyethylrutoside 2%
- Coenzyme Q_{10} 1% + asiaticoside 1%
- Coenzyme Q_{10} 1% + tribenoside 1%
- Coenzyme Q_{10} 1% + delphinidin 1%.

Ointments

- Coenzyme Q_{10} 1% + escin 1%
- Coenzyme Q_{10} 1% + sodium heparin 10,000 IU/100 g of ointment
- Coenzyme Q_{10} 1% + natural heparinoid 70,000 IU/g
- Coenzyme Q_{10} 1% + sulfate glucuronylglycosaminoglicane (heparinoid) 200 mg, equal to 400 UHC.

Suppositories

- Coenzyme Q_{10} 50 mg + escin 50 mg
- Coenzyme Q_{10} 100 mg + hydroxyethylrutoside 500 mg
- Coenzyme Q_{10} 50 mg + total extract of Centella asiatica (equal to 10 mg of asiaticoside) 50 mg
- Coenzyme Q_{10} 100 mg + tribenoside 1,000 mg
- Coenzyme Q_{10} 100 mg + delphinidin 400 mg.

The compositions of the invention may suitably be administered 2 to 4 times daily, even for prolonged treatment, at dosages depending on age, weight and general conditions of the patient.

CLAIMS

1. A pharmaceutical composition having vasculoprotecting activity, containing a ubiquinone compound and at least one other compound having known vasculoprotecting activity.

2. A pharmaceutical composition as claimed in claim 1, wherein the ubiquinone compound is Coenzyme Q_{10}.

3. A pharmaceutical composition as claimed in claim 1 or 2, wherein the known compound having vasculoprotecting activity is selected from compounds having flavanoid, heparinoid, terpenic or glycosidic structure.

4. A pharmaceutical composition as claimed in claim 3, wherein the compound having known vasal protecting activity is selected from the groups comprising: rutin, 0-(β-hydroxy-ethylrutoside), delphinidin, hesperidin, diosim, asiaticoside, catechin, heparin, heparinoids, escin, ruscogenin, tribenoside, sodium 2-phenyl-5,7-dioxyacetate and benzo-γ-pyrone.

5. A pharmaceutical composition as claimed in any one of claims 1 to 4, wherein Coenzyme Q_{10} and vasal protecting compound are present in a ratio ranging from 1:1 to 1:1000.

6. A pharmaceutical composition as claimed in any one of claims 1 to 5, for oral, parenteral, rectal or

topical administration.

7. A pharmaceutical composition as claimed in claim 6, in the form of capsules, tablets, syrup, granulate, sterile solution, ointment, cream, lotion, gel or suppositories.

8. A pharmaceutical composition substantially as described herein and exemplified.

9. A pharmaceutical composition according to any one of the preceding claims for use in vascular therapy.

Printed in the UK for HMSO, D8818935, 9/86, 7102.
Published by The Patent Office, 25 Southampton Buildings, London, WC2A 1AY, from which copies may be obtained.